

Application of Robert Getts
Serial No. 09/802,162 filed 3/8/2001
Response of 9/11/08 to Office Action of 3/11/08

Listing of Claims

This listing of claims will replace all prior versions and listings of claims in the application:

Listing of Claims

1. (Previously Presented) A method for detection and assay on a microarray, said method comprising the steps of:
 - 1) taking a microarray having thereon a plurality of features each comprising a first particular first nucleotide sequence;
 - 2) taking a first component comprising cDNA reagents having a capture sequence; and taking a second component comprising a dendrimer having at least one first arm comprising a label and at least one second arm having a second nucleotide sequence;wherein said cDNA reagents comprise a plurality of different nucleotide sequences, and wherein said capture sequence of said cDNA reagents is a common sequence among said cDNA reagents, said common sequence being complementary to said second nucleotide sequence of said dendrimer, said capture sequence being used for binding of said dendrimers to said cDNA reagents, such that said second arm of said dendrimer can bind to any of said cDNA reagents having said capture sequence by hybridization of said second nucleotide sequence of said dendrimer to said capture sequence;

Application of Robert Getts
Serial No. 09/802,162 filed 3/8/2001
Response of 9/11/08 to Office Action of 3/11/08

- 3) mixing said first and second components at a temperature and for a time sufficient to enable said first component to bind to said second component; and
 - 4) incubating this mixture with said microarray to enable the first nucleotide sequence to bind to said first component, wherein such binding results in the generation of a hybridization pattern on the microarray;
 - 5) wherein said first component comprising cDNA reagents is simultaneously hybridized to both said microarray and to said second component comprising dendrimer, while said first component comprising cDNA is on said microarray.
2. (Previously Presented) The method of claim 1, wherein said cDNA reagents are obtained from mRNA of a target sample, and further comprising the step of forming the first component comprising the cDNA reagents by contacting the mRNA with a quantity of a RT primer having the capture sequence, and with a reverse transcriptase, and nucleotide under conditions sufficient for initiating reverse transcription of the mRNA into said cDNA reagents.
 3. (Previously Presented) The method of claim 2 further comprising the step of purging excess unhybridized RT primer from said first component prior to incubation of said mixture.

Application of Robert Getts

Serial No. 09/802,162 filed 3/8/2001

Response of 9/11/08 to Office Action of 3/11/08

4. (Previously Presented) The method of claim 3 wherein the purging step further comprises the step of passing the first component through a spin column media to remove excess RT primer.
5. (Previously Presented) The method of claim 1 wherein the temperature sufficient to enable the second component to bind to the first component is from about 50 to 55°C.
6. (Previously Presented) The method of claim 1 wherein the temperature sufficient to enable the first component to bind to the first nucleotide sequence is from 42 to 65°C.
7. (Previously Presented) The method of claim 1 wherein the temperature sufficient to enable the first component to bind to the first nucleotide sequence is from about 4 to greater than 72 hours.
8. (Previously Presented) The method of claim 1 wherein the time sufficient to enable the second component to bind to the first component is from about 0.25 to 1 hour.
9. (Previously Presented) The method of claim 1, wherein the microarray and the mixture are incubated overnight at the temperature from about 42 to 65°C in a humidified chamber.
10. (Previously Presented) The method of claim 1, further comprising scanning the microarray

Application of Robert Geiss
Serial No. 09/802,162 filed 3/8/2001
Response of 9/11/08 to Office Action of 3/11/08

for detecting the detectable signal and the hybridization pattern generated.

11. (Previously Presented) The method of claim 1, further comprising washing the microarray to purge dendrimers unattached to microarray after the incubation of the microarray and the mixture.
12. (Previously Presented) The method of claim 11, wherein the washing step further comprises:
washing the microarray with 2X SSC buffer containing 0.2% SDS at 55°C for about 10 minutes;
washing the microarray with 2X SSC buffer at about room temperature for about 10 minutes;
and
washing the microarray with 0.2X SSC buffer at about room temperature for about 10 minutes.
13. (Previously Presented) The method of claim 1, wherein the mixture further comprising a hybridization buffer.
14. (Previously Presented) The method of claim 13, wherein the hybridization buffer further comprising 0.25 M NaPO₄, 4.5% SDS, 1 mM EDTA, and 1X SSC.
15. (Previously Presented) The method of claim 13 wherein the hybridization buffer further

Application of Robert Getts
Serial No. 09/802,162 filed 3/8/2001
Response of 9/11/08 to Office Action of 3/11/08

comprising 40% formamide, 4X SSC, and 1% SDS.

16. (Previously Presented) The method of claim 3 wherein the purging step further comprises the use of a hybridization chamber.
17. (Previously Presented) The method of claim 3 wherein the purging step further comprises the use of a hybridization station.
18. (Previously Presented) A method for detection and assay on a microarray, said method comprising the steps of:
 - 1) incubating a mixture including:
 - a first component comprising cDNA reagents having a capture sequence; and
 - a second component comprising a dendrimer having at least one first arm comprising a label and at least one second arm having a second nucleotide sequence;

wherein said cDNA reagents comprise a plurality of different nucleotide sequences, and wherein said capture sequence of said cDNA reagents is a common sequence among said cDNA reagents, said common sequence being complementary to said second nucleotide sequence of said dendrimer, said capture sequence being used for binding of said dendrimers to said cDNA reagents, such that said second arm of said dendrimer can bind to any of said cDNA reagents having said capture sequence by

Application of Robert Getts
Serial No. 09/802,162 filed 3/8/2001
Response of 9/11/08 to Office Action of 3/11/08

hybridization of said second nucleotide sequence of said dendrimer to said capture sequence;

said incubation being conducted at a first temperature and for a time sufficient to induce said first component to bind to said second component to form a prehybridized cDNA-dendrimer complex;

- 2) contacting a microarray having thereon a plurality of features each comprising a particular first nucleotide sequence with said mixture; and
 - 3) incubating said microarray and said prehybridized cDNA-dendrimer complex at a second temperature and for a time sufficient to induce said prehybridized cDNA-dendrimer complex to bind to said first nucleotide sequence, wherein such binding results in the generation of a hybridization pattern on said microarray.
19. (Previously Presented) The method of Claim 18, wherein said cDNA reagents are obtained using a spin column which is used to remove RT primer.
20. (Previously Presented) The method of Claim 1, wherein said mixing of said first and second components is conducted on said microarray.

Application of Robert Getts
Serial No. 09/802,162 filed 3/8/2001
Response of 9/11/08 to Office Action of 3/11/08

21. (Previously Presented) The method of Claim 1, wherein said mixing of said first and second components is conducted off of said microarray.
22. (Previously Presented) The method of claim 18, wherein said cDNA reagents are obtained from mRNA of a target sample, and further comprising the step of forming the first component comprising the cDNA reagents by contacting the mRNA with a quantity of a RT primer having the capture sequence, and with a reverse transcriptase, and nucleotide under conditions sufficient for initiating reverse transcription of the mRNA into said cDNA reagents.
23. (Previously Presented) The method of claim 22, further comprising the step of purging excess unhybridized RT primer from said first component.
24. (Previously Presented) The method of claim 23, wherein the purging step further comprises the step of passing the first component through a spin column to remove excess RT primer.
25. (Previously Presented) The method of claim 23, wherein the purging step further comprises the use of a hybridization chamber.
26. (Previously Presented) The method of claim 23, wherein the purging step further comprises the use of a hybridization station.

Application of Robert Getts
Serial No. 09/802,162 filed 3/8/2001
Response of 9/11/08 to Office Action of 3/11/08

27. (Previously Presented) A method as claimed in claim 1, wherein said method comprises an assay using dual channel analysis, said dual channel analysis comprising reverse transcription of two different capture sequences.
28. (Previously Presented) A method as claimed in claim 1, wherein said said method comprises an assay using multiple channel analysis, said multiple channel analysis comprising reverse transcription of at least three different capture sequences.
29. (Previously Presented) A method as claimed in claim 18, wherein said method comprises an assay using dual channel analysis, said dual channel analysis comprising reverse transcription of two different capture sequences.
30. (Previously Presented) A method as claimed in claim 18, wherein said said method comprises an assay using multiple channel analysis, said multiple channel analysis comprising reverse transcription of at least three different capture sequences.
31. (Previously Presented) A method as claimed in claim 2, wherein said method comprises an assay using dual channel analysis, said dual channel analysis comprising reverse transcription of two different capture sequences.
32. (Previously Presented) A method as claimed in claim 2, wherein said said method comprises

Application of Robert Getts
Serial No. 09/802,162 filed 3/8/2001
Response of 9/11/08 to Office Action of 3/11/08

an assay using multiple channel analysis, said multiple channel analysis comprising reverse transcription of at least three different capture sequences.

33. (Previously Presented) A method as claimed in claim 22, wherein said method comprises an assay using dual channel analysis, said dual channel analysis comprising reverse transcription of two different capture sequences.
34. (Previously Presented) A method as claimed in claim 22, wherein said said method comprises an assay using multiple channel analysis, said multiple channel analysis comprising reverse transcription of at least three different capture sequences.
35. (Previously Presented) A method as claimed in claim 1, wherein said capture sequence comprises more than one type of base.
36. (Previously Presented) A method as claimed in claim 1, wherein said capture sequence comprises adenine, guanine, cytosine and thymine bases.
37. (Previously Presented) A method as claimed in claim 18, wherein said capture sequence comprises more than one type of base.
38. (Previously Presented) A method as claimed in claim 18, wherein said capture sequence

Application of Robert Getts
Serial No. 09/802,162 filed 3/8/2001
Response of 9/11/08 to Office Action of 3/11/08

comprises adenine, guanine, cytosine and thymine bases.

39. (Previously Presented) A method as claimed in claim 1, wherein said method is used for expression analysis.
40. (Previously Presented) A method as claimed in claim 18, wherein said method is used for expression analysis.
41. (Previously Presented) A method as claimed in claim 1, wherein said cDNA reagents comprise a sample, and wherein said method is conducted to achieve hybridization of said dendrimer to said capture sequence such that each positive signal on said microarray can be counted to obtain quantitative information about the genetic profile of said sample.
42. (Previously Presented) A method as claimed in claim 18, wherein said cDNA reagents comprise a sample, and wherein said method is conducted to achieve hybridization of said dendrimer to said capture sequence such that each positive signal on said microarray can be counted to obtain quantitative information about the genetic profile of said sample.